The three infectious *Banana streak virus* species present in the banana genome of Pisang Klutuk Wulung (PKW) are allelic *

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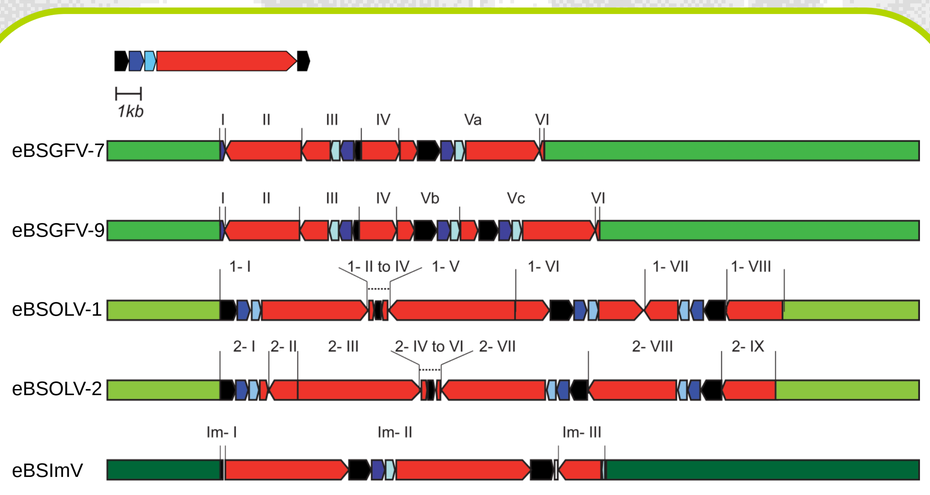
Context: The genome of banana (*Musa* sp.) harbours multiple integrations of *Banana streak virus* (eBSV), whereas this badnavirus does not require integration for the replication of its ds DNA genome. Some eBSV, only existing in the *Musa balbisiana* genome, are infectious by releasing a functional viral genome following stresses such as those existing in *in vitro* culture and interspecific crosses context. The structure of these eBSV is much longer than a single BSV genome, composed of viral fragments duplicated and more or less extensively rearranged (Gayral et al., 2008; Iskra-Caruana et al., 2010).

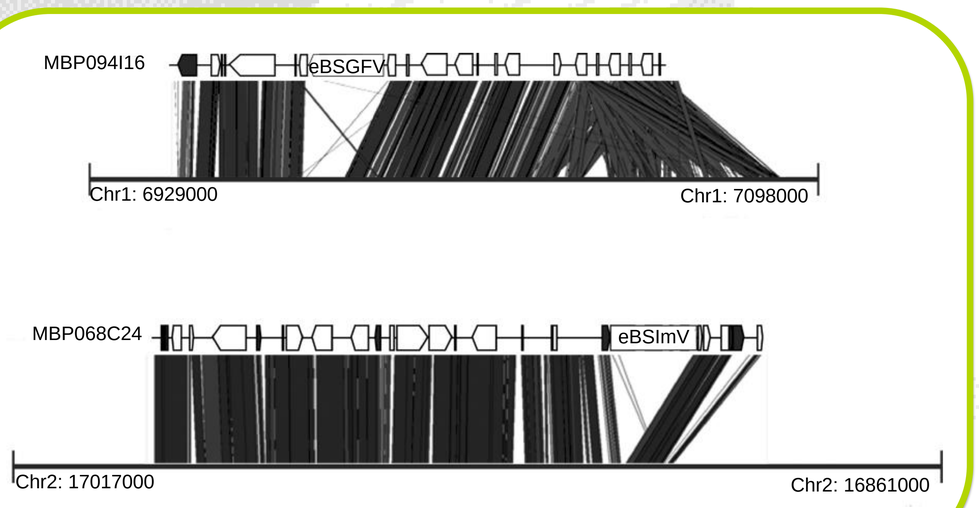
Seedy *M. balbisiana* diploid genotypes (BB) such as Pisang Klutuk Wulung (PKW) harbour such infectious eBSV belonging to three widespread species of BSV (*Goldfinger* -BSGFV, *Imové* – BSImV and *Obino l'Ewai* - BSOLV). The study of eBSV segregation among an AAB F1 progeny from the interspecific genetic cross involving PKW (BB) as the female parent and cv. IDN 110T (AAAA) as the male parent has revealed that integrated BSV species are allelic. We confirmed by bio-informatic analyses and fluorescent *in situ* hybridization that all three eBSV species are allelic and that eBSGFV and eBSOLV co-localise on chromosome 1 of PKW whereas eBSImV is on chromosome 2.

The wild diploïd *Musa balbisiana* (BB) Pisang klutuk wulung (PKW)



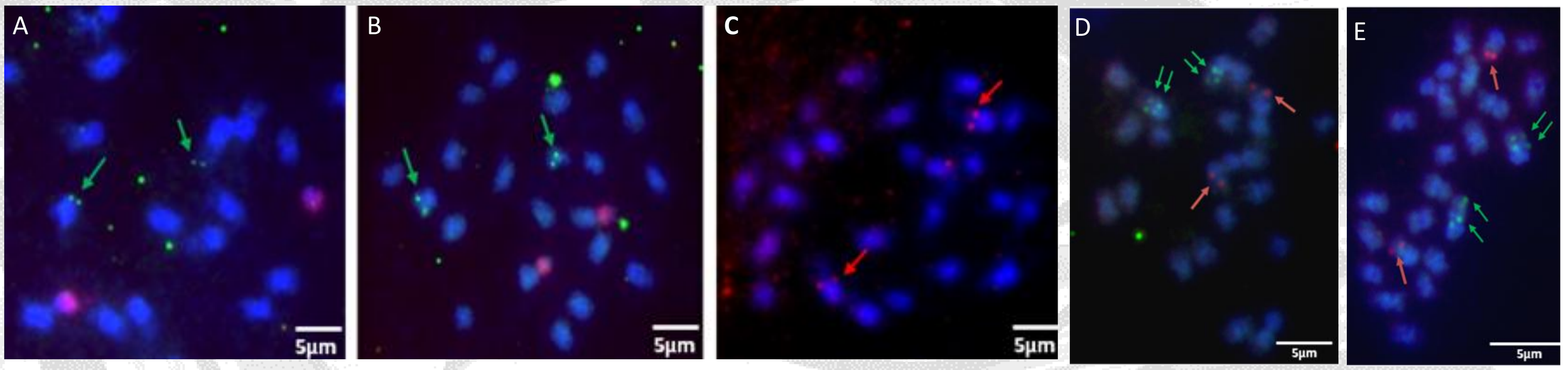
Molecular structures of eBSV in the diploid *Musa balbisiana* (BB) *Pisang klutuk wulung* (PKW):

- eBSGFV = Integrated sequence of *Banana streak Goldfinger virus*
- eBSOLV = Integrated sequence of *Banana streak Obino l'Ewai virus*
- eBSImV = Integrated sequence of *Banana streak Imové virus*



eBSV locus synteny with the *Musa acuminata* reference genome (D'Hont et al. 2012).

MBP094I16 and MBP068C24 refer to BACs containing eBSGFV and eBSImV, respectively. Strong similarities were found for BACs containing eBSGFV with chromosome 1 and for BACs containing eBSImV with chromosome 2. No match was found for BAC containing eBSOLV, probably due to the high TE content of the BAC clone.



eBSV localisation by Fluorescent *In Situ* Hybridization (FISH): Hybridizations (adapted from D'Hont et al., 2000) were performed using full-length genome of each BSV species as probe. Labelling was done by random priming with digoxigenine, biotine or Alexa 555.
Panels A and B: green signals correspond to BSGFV and BSImV respectivly and pink diffuse signals correspond to the 45S rDNA sequence.
Panel C: red signals correspond to BSLOV.
Panels D and E: green signals correspond to BSGVFV and BSOLV when red signals correspond to BSImV.
Chromosomes are counterstained with DAPI.

Conclusion: FISH result have undoubtedly confirmed the bio-informatic data and shown that eBSGVFV and eBSImV were located in two different chromosomes in the seedy diploïd *Musa balbisiana* (BB) PKW. Interestingly, they also clearly indicated that eBSOLV and eBSGFV co-localised on chromosome 1 and that all three eBSV species are allelic. This work should contribute to develop in a near future strategies aiming at reintroducing B genome in banana breeding programs without activable eBSV .

References:
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